

Sulfation of polysaccharides by Sodium Pyrosulfate in Dimethyl Sulfoxide

Overview :

The β -glucan (Laminarin) and its sulfated derivative (Laminarin Sulfates) have been previously demonstrated to induce resistance in grapevine against downy mildew (*Plasmopara viticola*).

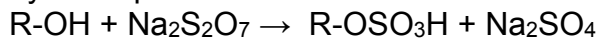
In plants, the fact that oligosaccharides must carry crucial sulfates for their biological function suggests that chemical sulfation of oligosaccharides can improve their biological properties.

In recent studies, compared to Laminarin (β -glucan), its sulfated derivative triggered an enhanced immunity against *P. viticola* in *V. vinifera* and a stronger immunity against TMV in *Nicotiana tabacum*. The results indicate that the chemical modification of an elicitor, such as sulfated derivative of β -glucan, could improve its resistance-inducer efficiency.

Moreover, if β -glucan is a substrate for plant β -1,3 glucanase, its sulfation clearly protects the molecule from its enzymatic degradation. Thus, a basal activity of plant glucanases can degrade β -glucan and consequently releases short inactive β -glucans whereas sulfated derivative still remains an active molecule during a longer period. This might explain the higher resistance induced by β -glucan sulfate compared to β -glucan.

So we tried to make glucan sulfates by the method of sulfation of polysaccharides with sodium pyrosulfate to improve their efficiency against downy mildew.

The sulfation of polysaccharides with sodium pyrosulfate can be schematically described by the equation :



The process is accompanied by the side reaction :



Sodium Pyrosulfate : $\text{Na}_2\text{S}_2\text{O}_7$

Dimethyl Sulfoxide = DMSO : $(\text{CH}_3)_2\text{SO}$

Protocol :

The sulfation of polysaccharides in the $\text{Na}_2\text{S}_2\text{O}_7$ -DMSO system can be performed following the steps above :

1. 1-5 g of a polysaccharide dried to constant weight, then, place it in a flask
2. Add Sodium pyrosulfate and dimethyl sulfoxide (pure grade) in succession with stirring, and heat the mixture for 6 h in a double boiler (water bath) at a temperature of 333 K. Composition of the sulfating mixture (g g^{-1}) : Curdlan : $\text{Na}_2\text{S}_2\text{O}_7$: DMSO = 1.0 : 3.0 : 5.0
3. After the lapse of the required time, neutralize the mixture with a saturated sodium carbonate solution to a pH between 8 and 10.
4. Then demineralize the solution by dialysis on cellophane membranes until the qualitative reaction for sulfate ions in wash waters was negative.
5. Put the purified solution in a refrigerator

Analyses :

The sulfur content of polysaccharide sulfates can be determined by potentiometric titration.
The Sulfo-Curdlan synthesized can be studied by IR spectroscopy.